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THE POLYCULTURE OF *Penaeus stylirostris* STIMPSON AND *Penaeus aztecus* IN TANKS

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ABSTRACT

Penaeus aztecus, obtained from the wild, and hatchery-reared *P. sty-tris* were placed together in 65 liter tanks in 5 ratios (0:100, 25: 0:50, 75:25 and 100:0). Two experiments were done. The first ex-ent consisted of 4 replicates of each ratio at 20°C and 30°C and 2 cates of each of the 100% tanks at 25°C. Initial size of *P. aztecus* . *stylirostris* were 0.12 and 0.07 g, respectively. In the second iment initial sizes of the wild *P. aztecus* and pond-reared *P. styli-is* were 1.30 and 0.89 g, respectively. The second experiment con-d of 4 replicates of each ratio at 25°C. The mean weight gain, mor-y and biomass (actual and that expected on the basis of monoculture) calculated.

In the first experiment, the 50% treatment had the highest mean t gain for both species. In both experiments, decreasing numbers *aztecus* (increasing numbers of *P. stylirostris*) in a treatment re-d in a better growth of *P. aztecus* in the treatment. The survival th species decreased as the percentage of *P. aztecus* increased. val was influenced by temperature, density, and species interac-. The largest difference between actual and expected biomass was to be for *P. aztecus* at 30°C, notably in the 75% *P. aztecus* treat-

P. aztecus does not seem to be influenced by the presence of *P. sty-tris* as much as *P. stylirostris* is influenced by *P. aztecus* in the tank.

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INTRODUCTION

Polyculture is the culture of two or more species together in the same facility. The biological basis of this concept calls for more efficient use both of a facility's environment and of available foods by stocking different species of organisms with varying behavior and feeding habits (Rabanal 1963). This idea of mixed culture can be found in many areas of the world including Asia and the Far East (Rabanal 1963; Villaluz et al. 1970; Liao 1977; Ling 1977; Tal Ziv 1978), the Middle East, Russia and the Mediterranean area (Yashouv 1966), and the United States (Fielding 1966; Bardach et al. 1972; Lovell 1979).

The first organisms purposely cultured together were various species of fish (Rabanal 1963). Penaeid shrimp have been cultured along with fish such as pompano (Tatum and Trimble 1978; Trimble 1980), mullet and catfish (Silva et al. 1977) and tilapia (Gundermann and Popper 1977). The polyculture of several species of shrimp usually has occurred by chance in ponds (Lunz 1951; Parker and Holcomb 1973) and in rice fields (Wickins 1976). Controlled polyculture in ponds has been done using *P. japonicus*, *P. semisulcatus*, *P. monodon*, and *Metapenaeus monoceros* (Lee and Liao 1970), *P. indicus*, *M. dobsoni*, *M. monoceros* and *M. affinis* (George 1975), *P. merguiensis*, *P. japonicus* and *P. monodon* (Gundermann and Popper 1977) and *P. stylirostris* and *P. vannamei* (Chamberlain et al. 1981). There has been only one previous report of tank polyculture of penaeid shrimps, with *P. monodon* and *P. penicillatus* in Taiwan (Liao 1977).

Tanks were used in this study to permit observation both of competitive behavior and species interactions (e.g., cannibalism, day-night activity, response to food) that cannot be determined directly during pond studies. Experimental temperatures were chosen to include values that might affect survival as well as growth for the species tested. The study thus assessed the reciprocal influences of the two species *P. aztecus* and *P. stylirostris* by evaluating combinations of the two in terms of factors such as increased weight and survival under the influence of controlled laboratory conditions in tanks.

MATERIALS AND METHODS

Aquarium tanks were placed in 3 temperature control rooms at the National Marine Fisheries Service (NMFS) Laboratory at Galveston, Texas. Each 75 x 32 x 31 cm tank was equipped with a "Eureka" undergravel filter covered by oyster shell and sand as described by Zein-Eldin (1963). These tanks were filled with filtered (5 micron cellulose filter) and sterilized (quartz ultraviolet light sterilizer) natural seawater from the adjacent Gulf of Mexico. The rooms had a cycle of 12 hours light and 12 hours dark and were set at 20, 25 and 30°C (±1°C) in the first experiment and 25°C (±1°C) in the second experiment.

Two sets of experiments were done using the native species *P. aztecus* and the non-indigenous species *P. stylirostris*. For the first experiment, *P. aztecus* were collected along the shore of a small bay near Galveston, Texas. The *P. stylirostris* (from Costa Rica) were hatched at the NMFS Galveston Laboratory, shipped to the Oceanic Institute for larval rearing (Waimanalo, Hawaii), and returned to the NMFS Laboratory for this experiment. Animals were blotted, weighed individually to the st 0.001 g initially and on day 14, 20 and 27 in experiment 1 and y 14 of the second experiment and returned to the tanks. The ini- mean weights of the animals were 0.12 g and 0.07 g for *P. aztecus* . *stylirostris*, respectively. Total biomass ranged from 1.4 g in containing 100% *P. stylirostris* to 2.4 g in tanks containing 100% *tecus*. The experiment was started on 23 April 1981 by stocking 20 p per tank and concluded on 22 May 1981 (30 days). The treatment nations consisted of 4 replicates of each of the following percen- ratios between species: 100:0, 75:25, 50:50, 25:75 and 0:100 at emperatures, 20 and 30°C. Also, there were two monoculture tanks ch species at 25°C.

The second experiment used *P. aztecus* collected from the same area Galveston, Texas, as in the previous experiment. The mean weight e *P. aztecus* was 1.30 g. The *P. stylirostris* (from Costa Rica) from females matured and spawned in captivity at the Texas A&M Uni- ty (TAMU) Shrimp Mariculture Facility (SMF), raised to postlarvae e Galveston Laboratory operated by TAMU and then raised to juve- (0.89 g) in ponds at Corpus Christi, Texas (TAMU SMF). The total ss ranged from 10.7 g in tanks containing 100% *P. stylirostris* to g in tanks containing 100% *P. aztecus*. At the start of this experi- on 27 May 1981, 12 animals were stocked in each of 20 tanks. Four cates were used for each of the 5 ratios of species used in the ous experiment. Temperature was held at 25°C (±1°C) and animals maintained 28 days until 23 June 1981.

The shrimp were fed ad libitum twice daily with feed developed for h and survival of shrimp in tanks (Fenucci and Zein-Eldin 1979). the growth (difference in mean weight and biomass) and percent val were determined. Expected final biomass was computed to exam- ossible species interactions based on measurements of survival and final size in monoculture.

Thus:

$$B_{et} = B_{ea} + B_{es}$$

and $B_{ea} = (N_a \text{ treatment})(S_a)(W_{fa})$

$B_{es} = (N_s \text{ treatment})(S_s)(W_{fs})$

here B_{et} = expected total biomass

B_{ea} = expected biomass from *P. aztecus*

B_{es} = expected biomass from *P. stylirostris*

and N_a = initial number of animals in treatment

S = % survival in monoculture

W_f = mean final weight in monoculture.

Weekly measurements were made of the temperature (hand-held mercury mometer) and salinity (optical refractometer) for each tank and were aged over the experimental period for each room. In experiment 1, al measured temperatures were 19.9°C (±0.5, S.E.M.), 25.3°C (±0.4) 30.6°C (±0.8). The salinity in experiment 1 was 27 ppt (±0.9). The al measured temperature and salinity in experiment 2 were 25.2°C 5) and 31 ppt (±0.6), respectively. There was no water exchange ng the entire experiment. Molts were counted and removed daily.

Statistical analysis was done on the combined data (weight gain, survival and biomass) of each replicate (tank) within a treatment using the Statistical Analysis System (SAS) programs (SAS Institute Inc., Cary, N.C.). Most data were analyzed using the contrasts procedure of the General Linear Models Procedure (GLM). Duncan's Multiple Range Test was used for additional analysis between treatments.

RESULTS AND DISCUSSION

MEAN WEIGHT GAIN AND GROWTH RATE

Mean weight gains at 20°C were 0.06, 0.08, 0.05 and 0.06 g for the 25, 50, 75 and 100% *P. stylirostris* treatments, respectively (Fig. 1). At 30°C, the gains for this species were 0.12, 0.19, 0.17 and 0.15 g for the 25, 50, 75 and 100% *P. stylirostris* treatments, respectively. Although the 50% *P. stylirostris* treatments had higher mean weight gains than other treatments at both temperatures, results of the contrast procedure (GLM) indicated no significant differences in mean weight gain when comparing ratios (P=0.1698) or ratio-temperature interactions. Overall, neither the presence of the second species nor its percentage in the treatment affected the mean weight gain of *P. stylirostris* (P=0.8607).

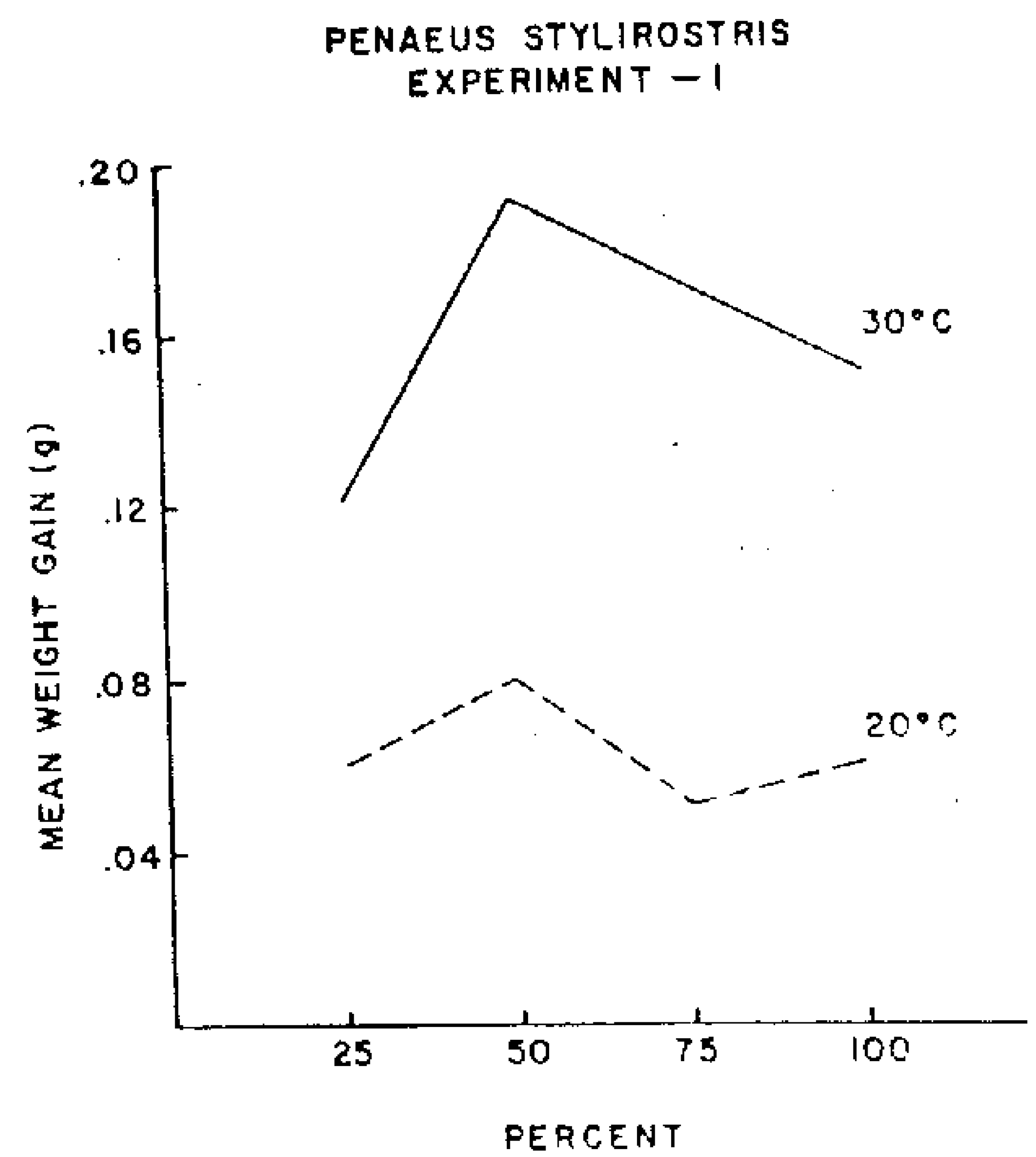
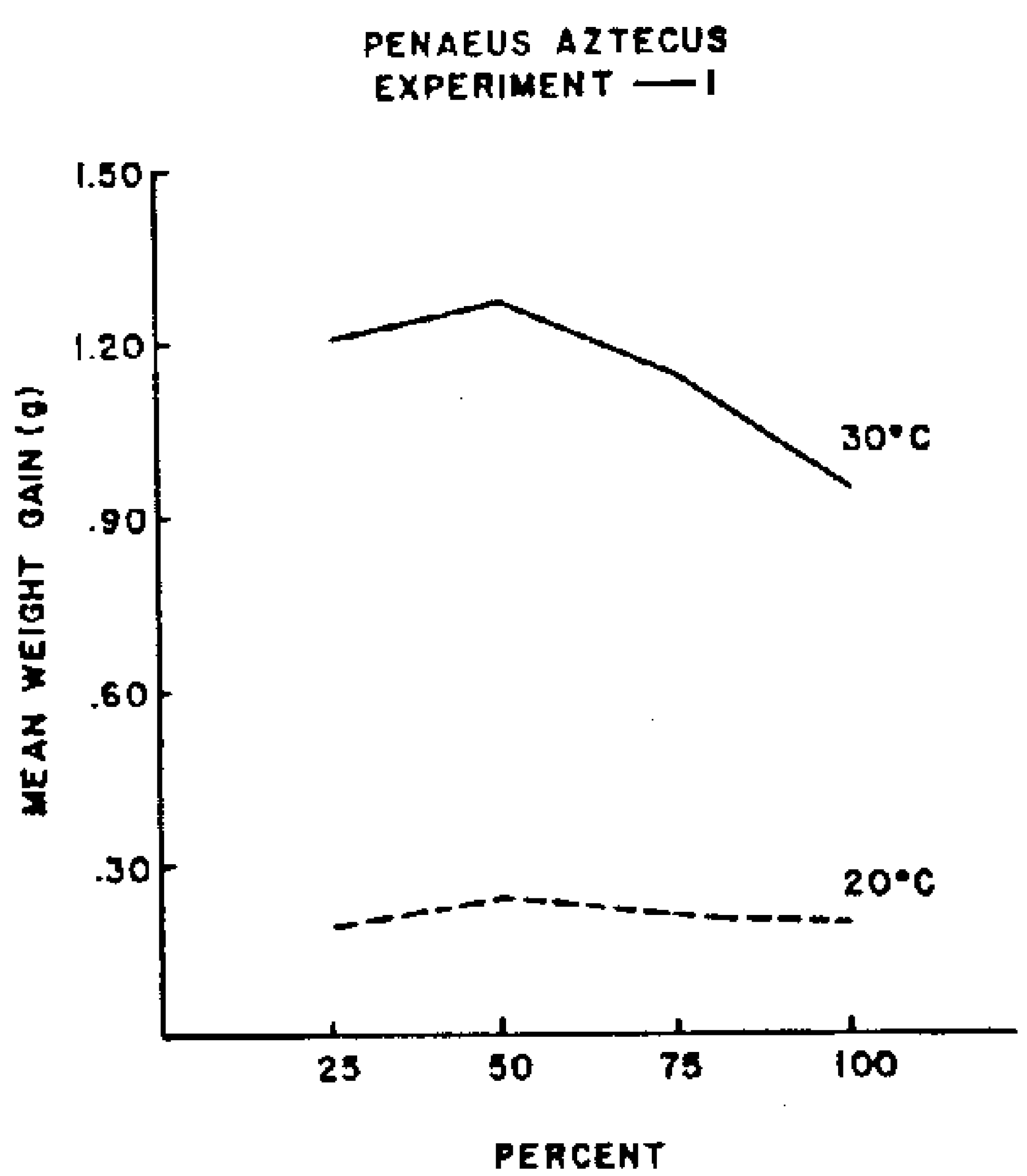


Figure 1. Mean weight of lab-hatched *Penaeus stylirostris* (0.07 g initial weight) held 30 days at 20°C and 30°C in the first experiment.

The initial mean weight of *P. stylirostris* was 0.07 g and the mean ght at the conclusion was 0.13 g at 20°C, 0.14 g at 25°C (two mono- ture tanks) and 0.22 g at 30°C, resulting in daily growth rates of 04 g/day at 20°C and 25°C, and 0.007 g/day at 30°C. The better wth (P=0.0001) at 30°C was expected since the optimum temperature for s species in the wild has been reported to range from 27 to 32°C nz and Bowers 1980). However, tank growth rates are much slower than se reported for pond growth (0.06 g/day at 27°C in nursery ponds) of *stylirostris* cultured with *P. vannamei* (Chamberlain et al. 1981).

P. aztecus did show some difference in mean weight gain between oculture and polyculture treatments in the first experiment. Mean ght gains in the individual treatment combinations were 0.19, 0.24, 2 and 0.20 g at 20°C, and 1.21, 1.27, 1.15 and 0.94 g at 30°C for the 50, 75 and 100% *P. aztecus* treatments, respectively. In this experi- t growth of *P. aztecus* may have been affected by *P. stylirostris* at C. The contrast procedure of GLM revealed that there was a signifi- t difference (P=0.005) in weight increase between *P. aztecus* alone when cultured with *P. stylirostris* at 30°C. It is not possible to ermine whether the growth of *P. aztecus* was positively affected by presence of *P. stylirostris* in high numbers (15-20 animals) or by a rease in numbers of *P. aztecus* (5-10 animals). At both temperatures highest mean weight gain for each species occurred in the 50% treat- t. There was a significant difference in mean weight gain within yculture treatments (P=0.02) at 30°C and Duncan's Multiple Range Test ealed that treatments containing only 25 and 50% *P. aztecus* in poly- ture yielded significantly higher weight gains (P=0.05) than treat- ts with 75 or 100% *P. aztecus*. At 20°C there were no significant dif- ences in weight gain of *P. aztecus* (Fig. 2) between monoculture and yculture treatments (P=0.35) or between ratios (P=0.43).



ure 2. Mean weight gain of wild *Penaeus aztecus* (0.12 g initial weight) held 30 days at 20 and 30°C in the first experiment.

As with *P. stylirostris*, *P. aztecus* grew larger at the warmer temperature. The initial mean weight of *P. aztecus* was 0.12 g while final means were 0.66 g at 20°C, 0.70 g at 25°C (two monoculture tanks) and 1.26 g at 30°C. These values represent daily growth rates of 0.02 g/day at 20 and 25°C, and 0.04 g/day at 30°C, resulting in a highly significant difference (P=0.0001) between the weights gained at 20 and 30°C. A similar growth rate at 30°C was also reported by Fenucci and Zein-Eldin (1979), using the same equipment, procedures and feed (0.44 g initial weight).

In the second experiment mean weight gains for *P. stylirostris* were 0.19, 0.19, 0.22 and 0.19 g in the 25, 50, 75 and 100% treatments, respectively (Fig. 3), while *P. aztecus* had mean weight gains of 1.31, 1.26, 1.04 and 1.00 g in the 25, 50, 75 and 100% treatments, respectively. At this one temperature (25°C), there were no statistically significant differences in mean weight gain between all polyculture treatments and monoculture of *P. stylirostris* (P=0.54) or *P. aztecus* (P=0.17). As in the first experiment, although 25 and 50% *P. aztecus* treatments had a higher mean weight gain than the 75 and 100% treatments, the treatment pairs were not significantly different (P=0.1393).

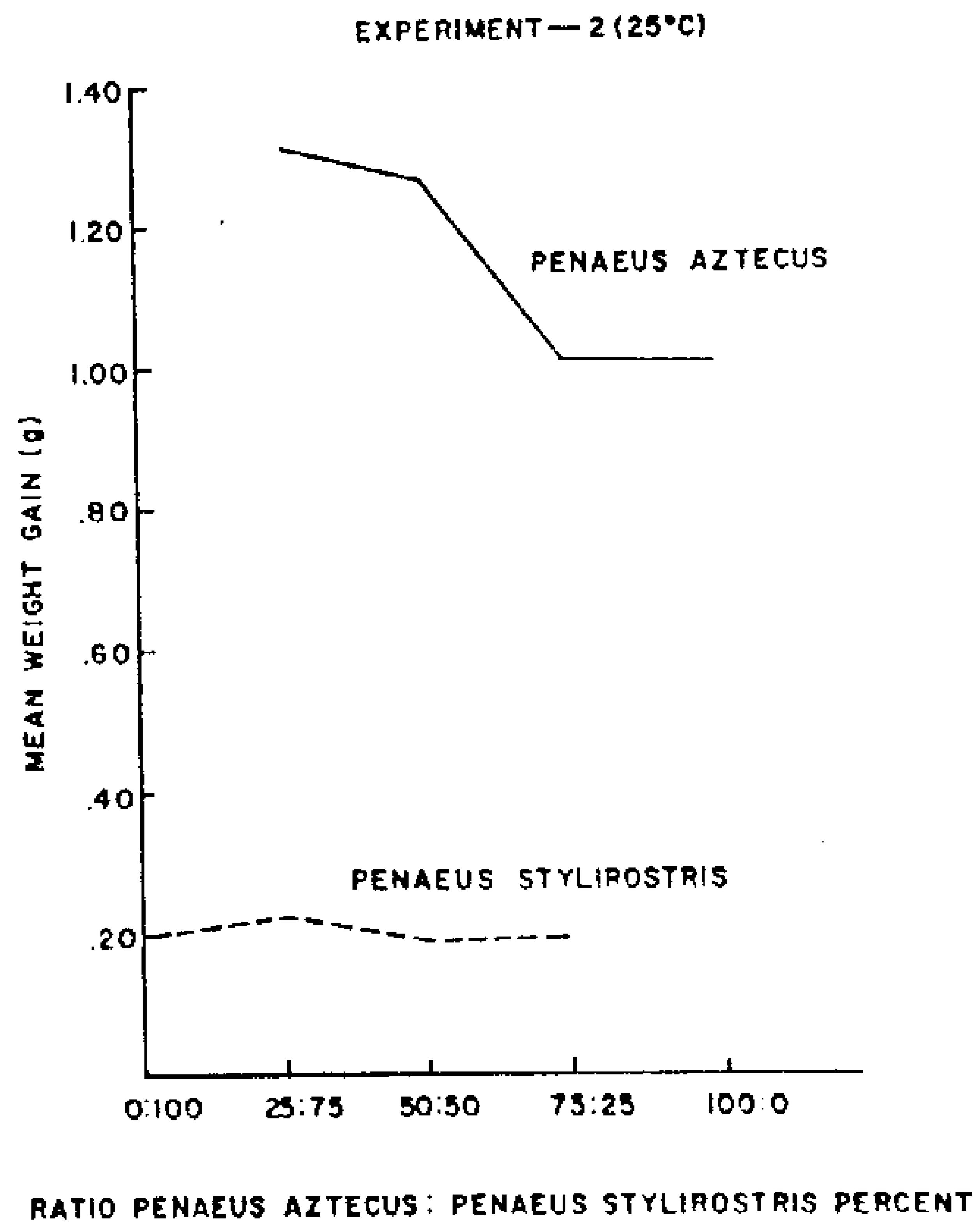
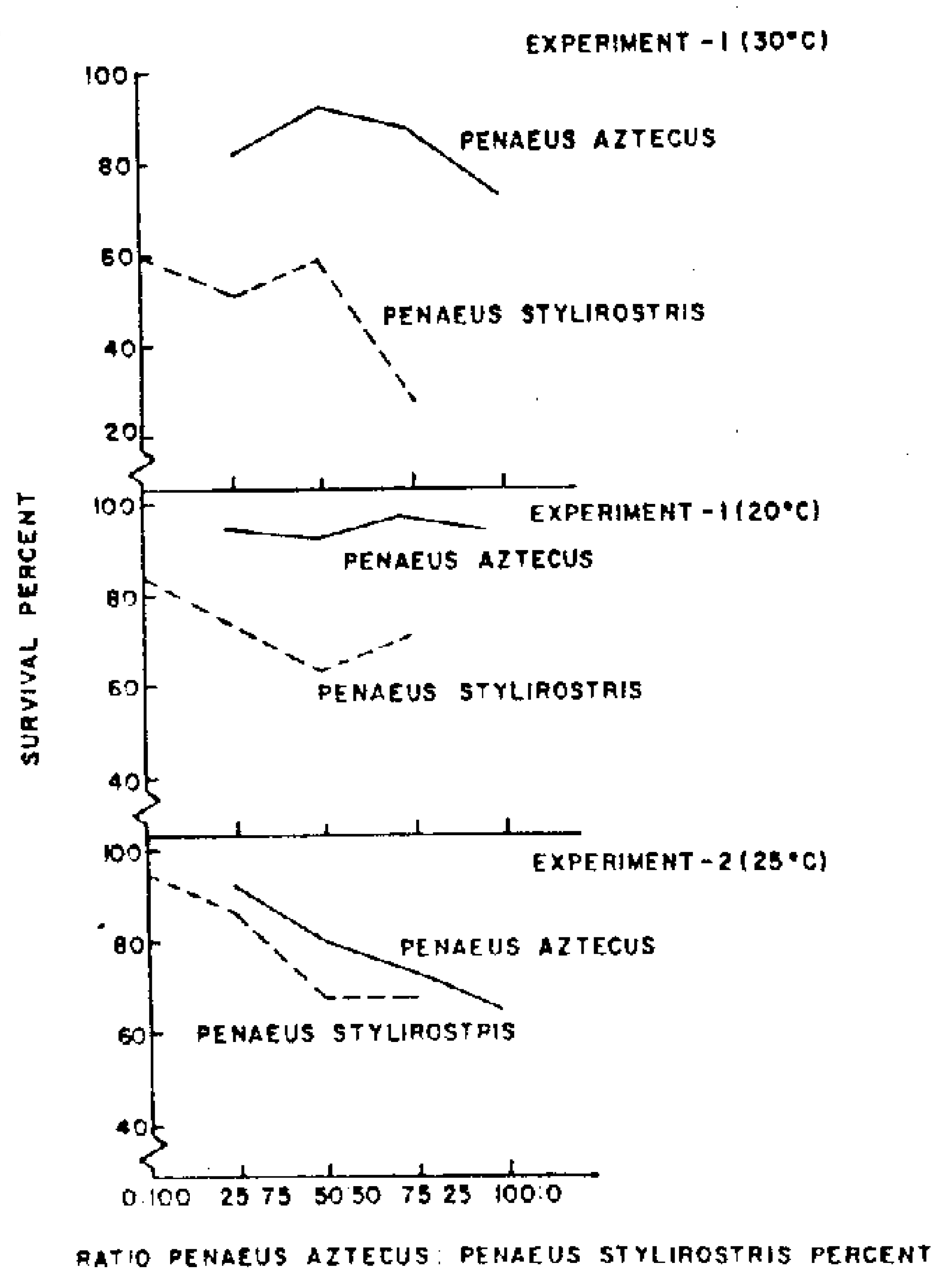


Figure 3. Mean weight gain of wild *Penaeus aztecus* (1.30 g initial weight) and pond-reared *P. stylirostris* (0.89 g initial weight) held 28 days at 25°C in the second experiment.

The initial mean weights were 0.89 and 1.30 g for *P. stylirostris* . *aztecus*, respectively. *P. stylirostris* had a final mean weight)8 g for a daily growth rate of 0.007 g. The mean weight for *P. us* after 28 days was 2.45 g, giving this species a growth of 0.04 . Growth rates for each species at 25°C were the same as that at in the first experiment, thus higher than at 25°C in the first ex- ent. Daily rate of growth of shrimp expressed as weight per day ds both upon temperature and initial size. Smaller animals usually e or triple in weight in a shorter period of time than larger ani- It thus appears that *P. aztecus* with its initial larger size r affected *P. stylirostris* negatively or that the group of cultured *ylirostris* had a much lower growth potential in tanks.

NT SURVIVAL

Percent survival varied with temperature in both species, with sur- better at 20°C than at 30°C. The overall survival rates of *P. sty- tris* were 72 and 48% at 20 and 30°C, respectively (Fig. 4). *P. az-* had higher survival rates of 95 and 81% at 20 and 30°C, respec- y, with a highly significant difference (P=0.0001) between survival at 20 and 30°C (*P. stylirostris* and *P. aztecus*). The survival of *tecus* at 30°C is similar to the 80% survival observed by Forster eard (1974) for this species in a tank (86 x 72 x 40 cm) experiment 8°C) after 28 days with 15 shrimp (0.3 g initial weight) per tank.



e 4. Mean survival after 30 days in experiment 1 (20 and 30°C) and after 28 days in experiment 2 (25°C).

There was an overall difference (P=0.02) of combined survival rates between treatments (ratios) in the warmer temperature. Within the 30°C temperature (Fig. 4), *P. stylirostris* survived best in the 100% treat- ment (60%) while only 25% survived in the 25% *P. stylirostris* treatment (Fig. 4). This low survival was due in part to one tank having no sur- vival of *P. stylirostris*. In contrast, 90% of the *P. aztecus* survived the 50% treatment, while the lowest survival (68%) was found in the 100% *P. aztecus* treatment (Fig. 4).

Within the second experiment at 25°C, survival among *P. aztecus* treatments at 25°C was not significantly different (P=0.1734), although survival among *P. stylirostris* treatments was significantly different (P=0.004). The overall survival rate for *P. stylirostris* (79%) and *P. aztecus* (76%) were similar (Fig. 4). The 100% *P. stylirostris* treatment had the highest survival of 96%, while the lowest survival for this spe- cies was 66% for the 25% (3 animals/tank) and 50% (6 animals/tank) *P. stylirostris* treatments. *P. aztecus* survived best in the 25% treatment (92%) and as in the earlier experiment, the lowest survival was in the 100% *P. aztecus* treatment (62%). *P. aztecus* survival was similar to the range of survival rates (80-100%) found by Zein-Eldin and Aldrich (1965) in tank experiments. Survival of both species generally decreased at all temperatures as the percentage of *P. aztecus* increased (Fig. 4), sug- gesting a species effect as well as density and biomass influences. Al- though total biomass/m^2 was greater and culture period shorter in this tank experiment than in ponds (the only data available for comparison), *P. stylirostris* survival exceed that reported for combinations of *P. sty- lirostris* with *P. vannamei* in ponds (Chamberlain et al. 1981).

BIOMASS

As expected, temperature affected mean biomass increase, resulting in highly significant differences (P=0.0001) between two temperature treatments (20 and 30°C; all ratios combined) for both species (Table 1). Although mean biomass increase within temperatures tended to increase with the percentage of that species in the treatment, paired comparisons showed that intermediate ratios were not significantly different for *P. stylirostris*.

The mean biomass increase per tank (within each treatment of each species) were 0.20, 0.50, 0.59 and 0.94 g at 20°C and 0.20, 1.10, 1.26 and 2.04 g at 30°C for the 25, 50, 75 and 100% *P. stylirostris* treat- ments, respectively. There was no significant difference between the 50 and 75% treatments (P=0.05) (Table 1).

The mean biomass increases per tank within each treatment were 3.47, 3.14, 2.21 and 0.91 g at 20°C and 12.71, 14.75, 11.39 and 4.90 g at 30°C for the 100, 75, 50 and 25% *P. aztecus* treatments, respectively. Be- cause of the relatively great mean biomass increase in the 75% treatment, there was no significant difference between the 75 and 100% *P. aztecus* treatments (P-0.05) at both temperatures (Table 1).

Comparisons of actual biomass with that expected on a basis of mono- culture indicated that *P. aztecus* in all combinations (except 25% at 20°C) grew better than in monoculture (Table 2). Conversely, biomass of *P. stylirostris* was less than expected in all combinations at 20°C, and clearly exceeded expectation only at 75% *P. stylirostris* at 25°C. Thus, the initially smaller *P. stylirostris* contributed more weight to the total biomass than expected only at 75% at 25°C.

1. Mean Biomass Increase per Tank for the First and Second Experiments. Total initial biomass per tank ranged from 1.4 g (*Penaeus stylirostris*) to 2.4 g (*P. aztecus*) in experiment 1. Total initial biomass per tank in experiment 2 ranged from 10.7 g (*P. stylirostris*) to 15.6 g (*P. aztecus*). Values with the same letter do not differ significantly (Duncan's Multiple Range Test, α = 0.05).

Treatment	Mean biomass increase (g)			
	Experiment 1		Experiment 2	
	20°C	25°C	30°C	25°C
P. aztecus (%)				
100	3.47^{a}	12.84	12.71^{a}	7.82^{a}
75	3.14^{a}	-	14.75^{a}	6.79^{b}
50	2.21^{b}	-	11.39^{b}	6.04^{b}
25	0.91^{c}	-	4.90^{c}	3.60^{c}
P. stylirostris (%)				
100	0.94^{a}	0.32*	2.04^{a}	1.74^{a}
75	0.59^{b}	-	1.26^{b}	1.76^{a}
50	0.50^{b}	-	1.10^{b}	1.03^{b}
25	0.20^{c}	-	0.20^{c}	0.53^{c}

42% survival.

:t 25°C mean biomass increases of the 75 and 100% *P. stylirostris* ·ents were approximately the same, while the other treatments were ·icantly different (P=0.05, Table 1). In contrast to the earlier ment, the mean biomass increases per tank for *P. aztecus* indicated ·icant differences between the 100 and 75% treatments, while 75 and ·eatments were similar (P=0.05).

'hen comparing the actual biomass with the calculated expected bio- ıt 25°C (Table 2), the actual biomass for both *P. aztecus* alone and ·e species combined was higher than expected for all treatments, ·he 50 and 25% *P. aztecus* treatments showing the largest difference ·n actual and expected biomass. The actual biomass of the *P. styli*-·*s* was less than expected in all but the 75% *P. stylirostris* treat- Thus, in both experiments, total actual biomass exceeded pre- ł biomass in almost all density and temperature combinations (Table ·his was primarily caused by *P. aztecus* which exceeded expected ·s in all conditions but 20°C and 25%, while *P. stylirostris* bio- ·as less than predicted at nearly all combinations.

CONCLUSION

·rowth of small *P. aztecus* appeared to be favorably influenced in ·esence of *P. stylirostris*, particularly in the 25 and 50% treat- . Whether the larger mean weight gain of *P. aztecus* (at low per- ·es) was actually related to the higher percentage of *P. styliros*- in the tank or to the decrease in biomass (decrease in number) of ·*ecus* could not be determined. The higher mean weight gain of *P.* ·*s* at lower ratios was statistically significant.

Table 2. Actual and Expected Biomass per Species Treatment (tanks combined) for Each Species Individually and in Combinations. Expected biomass calculated from biomass and survival of the species in monoculture.

Ratio (%)	Temperature (°C)																	
	20						30						25					
	P. az.		*P. styl.*		Comb.		*P. az.*		*P. styl.*		Comb.		*P. az.*		*P. styl.*		Comb.	
P. az.:*P. styl.*	act.	(ex.)	act.	(ex.)	act.	(ex.)	act.	(ex.)	act.	(ex.)	act.	(ex.)	act.	(ex.)	act.	(ex.)	act.	(ex.)
100:0	24.1	-	-	-	24.1	-	57.4	-	-	-	57.4	-	69.0	-	-	-	69.0	-
75:25	19.6	(18.1)	1.8	(2.1)	21.4	(20.3)	65.2	(43.4)	0.9	(2.9)	66.1	(46.3)	61.2	(51.8)	8.9	(11.0)	70.1	(62.8)
50:50	13.6	(12.1)	36	(4.3)	17.2	(16.4)	49.9	(28.7)	6.0	(5.9)	55.9	(34.6)	48.5	(34.5)	17.7	(22.0)	66.2	(56.5)
25:75	5.9	(6.0)	5.6	(6.4)	11.5	(12.4)	21.8	(14.4)	7.1	(6.8)	28.9	(21.2)	28.9	(17.3)	35.2	(33.1)	62.1	(50.4)
0:100	-	-	8.5	-	8.5	-	-	-	11.8	-	11.8	-	-	-	44.1	-	44.1	-

Survival was better for *P. aztecus* than *P. stylirostris*, with sur- greater than 92% of *P. aztecus* at the lower temperature (20°C) and ally greater than 78% at 30°C (Fig. 4). This species is normally in cooler waters, and postlarvae grow and survive well at 20-25°C -Eldin and Griffith 1969). *P. stylirostris* also had a higher sur- rate at the 20°C temperature (72%) than at the 30°C temperature even though *P. stylirostris* is considered a warm water species an optimum range of 27-32°C (Menz and Bowers 1980).

At 25°C, *P. stylirostris* had a slightly better overall survival (79%) than *P. aztecus* (76%). *P. stylirostris* survival improved increasing numbers of *P. stylirostris* (3-12 individuals) and de- ing numbers of *P. aztecus* (12-3 individuals) (Fig. 4). Thus, the *P. aztecus* in a treatment, the higher the mortality of both *P. sty- tris* and *P. aztecus*, the latter contributing almost two times the ss (12.9 g/tank) as *P. stylirostris* (7.04 g/tank). Although there possibility that *P. aztecus* cannibalized the smaller *P. styliros-*, there was no evidence of this.

Under the conditions of these experiments, the native *P. aztecus* rmed better in mean weight gain, survival and biomass increase than non-indigenous, hatchery-reared *P. stylirostris*. Overall, the in- se in weight of *P. aztecus* was 5 times at 20°C, 6 times at 25°C (2) and 10 times at 30°C. However, *P. stylirostris* doubled its weight) and 25°C and only tripled its weight at 30°C. *P. stylirostris* have been expected to produce higher growth rates (in terms of e in weight/unit body weight/unit time) at 30°C than *P. aztecus* be- smaller initial size of *P. stylirostris* should have resulted in an entially higher growth rate than the larger *P. aztecus*.

Survival of both species depended on the number of *P. aztecus* in treatment with low numbers of *P. aztecus* in the treatment resulting igher survival rates. The largest difference between actual and ex- d total biomass in a combination was for the 50% *P. aztecus*:50% *P. irostris* treatments. The largest differences between actual and ex- d total biomass were found for *P. aztecus* at 30°C. *P. aztecus* does seem to be influenced by the presence of *P. stylirostris* as much as *tylirostris* is influenced by *P. aztecus* in the same tank. Therefore, species are possible candidates for small polyculture systems such nks, based on the criterion of Brick and Stickney (1979) that the ndary species (*P. stylirostris*) did not adversely affect the primary ies (*P. aztecus*) but increased the total production in at least some itions.

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